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This invention provides a simple and convenient, single stage, single vessel cell extraction and assay method which is suitable for the extraction and measurement of a range of different types of analyte which occur as cellular components. The invention also provides kits of reagents suitable for performing cellular extraction and measurement as a single stage, single vessel process.

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In the Claims

Please amend claims 1-13, as set out below:

1. [Amended] A method of assaying for an analyte in a cell sample which method comprises the steps of:

i) mixing a sample of cells possibly containing the analyte with a cell lysis reagent to provide a [cell lysis fluid] lysed cellular sample;

ii) mixing the [cell lysis fluid] lysed cellular sample with assay reagents, including a specific binding partner of the analyte [for binding to the analyte, for performing a specific binding assay for the analyte,] to perform a specific binding assay by forming a reaction mixture comprising a specific binding partner-analyte complex;

iii) [and] mixing the [cell lysis fluid] lysed cellular sample with a sequestrant for the cell lysis reagent, whereby the specific binding assay of step ii) is performed in the presence of the sequestrant; and

iv) detecting the presence of the specific binding partner-analyte complex.

2. [Amended] [A] The method as claimed in claim 1, wherein the cell lysis reagent is a detergent.

3. [Amended] [A] The method as claimed in claim 1, wherein the sequestrant is a cyclodextrin.

4. [Amended] [A] The method as claimed in claim 3, wherein the amount of sequestrant is in the range of 1 - 5% of the [binding reaction mixture] said reaction mixture.

5. [Amended] [A] The method as claimed in claim 1, wherein steps i), ii) and iii) are all performed in a single reaction vessel.

6. [Amended] [A] The method as claimed in claim 1, wherein [multiple] individual assays are performed in parallel in individual vessels which are wells of a multiwell plate.

7. [Amended] [A] The method as claimed in claim [1] 5, wherein the cells are cultured in [a] said vessel and are lysed in that vessel for assaying the analyte in that vessel.

8. [Amended] [A] The method as claimed in claim 1, wherein the assay of step ii) is a homogenous assay.

9. [Amended] [A] The method as claimed in claim 1, wherein the assay of step ii) is a scintillation proximity assay.

10. [Amended][A] The method as claimed in claim 1, wherein the specific binding assay of step ii) is an immunoassay.

11. [Amended] [A] The method as claimed in claim 1, wherein the analyte is adenosine-3', 5'-cyclic monophosphate, the cell lysis reagent is dodecyl trimethyl ammonium bromide and the sequestrant is α -cyclodextrin.

12. [Amended] [A] The method as claimed in claim 1, wherein the cells have been maintained in a culture medium, and step i) is performed in the presence of the culture medium.

13. [Amended] [A] The method as claimed in claim 1, wherein the intracellular or [the total (intracellular plus extracellular)] both intracellular and extracellular concentration of the analyte is measured [of an] and the analyte is selected from the group consisting of adenosine-3',5'-cyclic monophosphate, interleukin-6 and prostaglandin E₂.